

The clinical relevance of antibody-mediated rejection: a new era of heart transplantation

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This issue reports a case where the authors can be congratulated on having saved the life of a patient who originally suffered from giant cell myocarditis and had been treated with cyclosporine (CyA), azathioprine (AZA) and corticosteroids (PDN) for 4 years before clinically deteriorating and been subjected to heart transplantation (HTx) [1]. The pretransplant work-up included human leukocyte antigen (HLA)-typing of both patient and donor, screening for anti-HLA antibodies and cross-match with regard to T- and B-cells being negative. Postoperatively, conventional immunosuppression (CyA, AZA and PDN) was introduced. There were three HLA Class I and one HLA Class II allele mismatches between donor and recipient.

The first endomyocardial biopsy (EMB) showed mild rejection (ISHLT 1R) without C4d deposition in immunohistochemistry. The second EMB (3 weeks postoperatively) revealed severe macrophage (CD68+), and eosinophilic and lymphocytic infiltration, again without C4d deposition. The patient developed fever without clinical signs of infection and was treated under the presumption of acute cellular rejection (ACR). After initial stabilization, the patient developed cardiogenic shock 2 days later, which required urgent treatment with extracorporeal membrane oxygenation, followed by the implantation of a biventricular assist device the next day. At that time, EMB showed severe ACR (ISHLT 3R) and antibody-mediated rejection (AMR), strongly positive for C4d [2, 3]. The cytotoxic T- and B-cell cross-match tests remained negative.

The endothelial precursor cells cross-match test (EPC-XM; XM-ONE®, AbSorber AB, Sweden) was synchronously performed with EMBs, measuring serum levels of the amount of antiendothelial-cell antibodies (AECAs), either against donor-specific EPCs or against autoreactive EPCs (the patient's own EPCs). After initially being negative, donor-specific AECA became positive at the first EMB (ISHLT 1R), but autoreactive AECAs were still negative. At the time of acute graft failure, the test was surprisingly positive for both donor-specific and autoreactive IgM antiendothelial AECA, but not for donor IgG AECA. The authors conclude that severe AMR was caused by non-HLA, AECAs. Epicrisis was successfully treated by plasmapheresis, intravenous immunoglobulin (IVIgG), rituximab, murenomab-CD3, bortezomib and re-HTx. One month after re-HTx, the patient had ACR (ISHLT 2R), but remained free of AMR.

During the last 15 years, AMR became a topic of major clinical relevance. Potential explanations are improved diagnostic tests and the rising population of patients having undergone VAD implantation before HTx. Its pathophysiology remains unknown to date.

AMR is often associated with haemodynamic compromise, increased mortality and development of graft coronary artery disease (CAD). Younger age, congenital heart disease, positive donor-specific cross-match, positive panel reactive antibody (PRA) titres, sensitization to OKT3, CMV seropositivity, previous blood transfusions and female gender have been identified as risk factors for AMR [4].

Ongoing discussions finally led to the consensus conference on April 20, 2010 in Chicago [5]. They also address the question of non-HLA antibodies, such as vimentin, endothelial cells, MICA/MICB, with a very low incidence (2/128) by using the current XM-One technique to separate EPCs from donor blood [6]. It was mentioned that 'more work is required to understand whether these assays can be used to diagnose AMR'.

The association of AMR with cardiovascular mortality, such as death resulting from acute rejection, myocardial infarction, congestive heart failure, graft failure, arrhythmia or CAD, was recently shown in paediatric HTx [7]. Such an observation supports protocol screening for AMR. However, the need for the treatment of measurable but clinically silent AMR and the algorithm to do so, still remains to be determined. Most of the transplant centres continue to perform event-driven and individualized strategies, as shown in the current case report.

Due to improved diagnostic testing and recently developed therapeutic strategies, we do have the chance to avoid or to treat AMR individualized and sufficiently, either in the case of preformed (such as in ABO incompatibility) or acquired HLA antibodies (such as via pregnancy, prior transfusion or transplant). Some may argue that most of the measures against AMR need intravenous administration, which is difficult to maintain over the years. However, we can assume that 'accommodation' will occur [8]. To provide an example of how to manage such cases successfully, we would like to share the case of accidental B to O incompatible HTx having been rescued successfully by anti-B antibody absorption, plasma exchange with AB fresh frozen plasma, IVIgG, by using an anti-B antibody

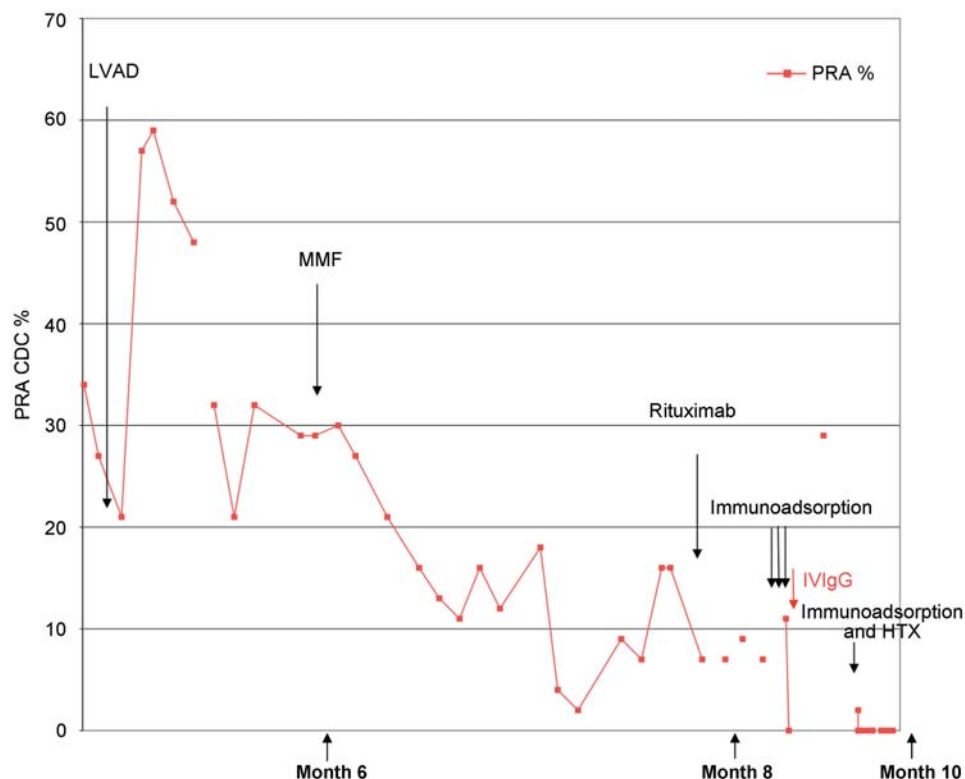


Figure 1: Desensitization protocol of the patient.

immunoabsorption column and the C1 esterase inhibitor (C1inh, Berinert®) [9]. The HTx patient survived 5 years and developed a histo-blood group type change of the graft from B to O [10].

Another example is a desensitization protocol of acquired anti-HLA Class I antibodies: a female patient underwent coronary artery bypass grafting. Ten years later, the patient received a gastric bypass due to adipositas permagna and colon resection due to diverticulitis. The patient had a previous pregnancy. Due to intra-abdominal bleeding with haemorrhagic shock, splenectomy was required. Prolonged perioperative hypotension led to an anterior myocardial infarction and congestive heart failure. As a consequence, left ventricular assist device (LVAD) implantation was required. Preoperative serum PRA levels were 43%, reaching postoperative serum PRA levels of 59%. In flow cytometry 10 days post-LVAD implantation, 21 different HLA Class I antibodies were detected. The virtual cross-match (etrl.eurotransplant.org/cms/index.php?page=services) yielded a very low likelihood for receiving a matching cardiac allograft. As desensitization protocol, we administered mycophenolic mofetil, rituximab and repetitive Protein-A immunoabsorption. After single rituximab administration, the absolute B-cell count dropped to <25/ μ l. Figure 1 depicts the desensitization protocol with a consecutive decrease of the PRA levels. The patient finally underwent HTx with two unfavourable matches (HLA-B8 and B27); we continued during the postoperative phase with Protein-A immunoabsorption procedures. The EMB 1 week after HTx showed slightly swollen endothelial cells, which led to the administration of IVIgG and C1inh. Figure 2 shows immunohistochemical staining of the heart biopsies. The EMB 2 weeks after HTx showed ACR (International Society of Heart and Lung Transplantation 2R), which was treated with methylprednisolone

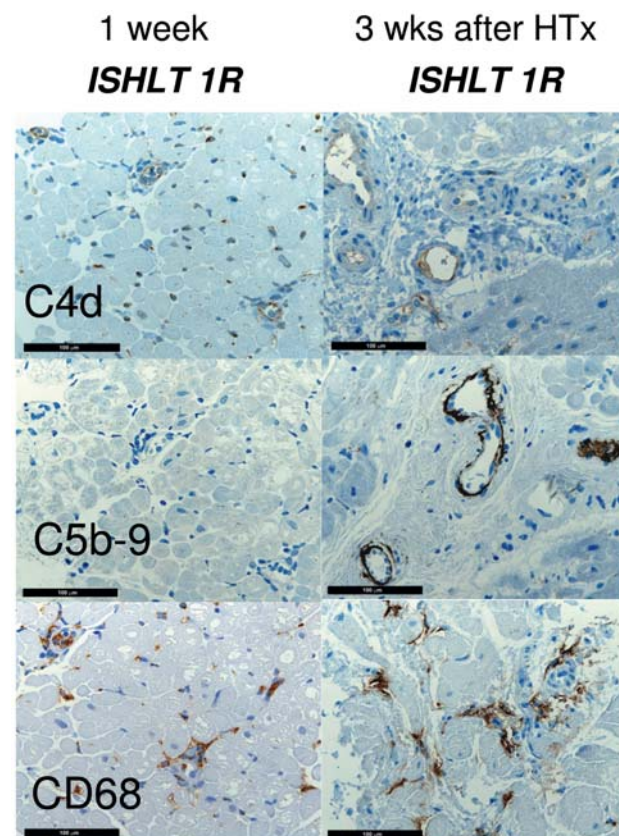


Figure 2: Immunohistochemical staining of heart biopsies: positive staining is highlighted by a brown colour. Detection of the classical complement pathway component C4d and terminal membrane attack complex C5b-9 as well as tissue macrophages by CD68.

and rATG-Fresenius. Six years after HTx, the patient continues to be fine.

The current case report by Sigurdardottir *et al.* and our examples show that AMR is a contemporary clinical challenge, which can be reasonably diagnosed, addressed and solved in an appropriate fashion.

One week after HTx (left panel) only minimal complement deposition was observed in small capillaries and larger vessels. This is an unspecific feature and may be observed in the immediate peritransplant period, e.g. following graft ischaemia/reperfusion. CD68 marks some scattered macrophages. Endothelial cells are flat and general cellularity is low. A minimal perivascular lymphocyte-predominant infiltrate suggests minor ACR.

Three weeks after transplantation (right panel), there is still only minimal C4d deposition, and slightly more C5b-9, however, mainly localized to larger blood vessels. Macrophage counts remain constant. The endothelial lining of only a few capillaries appears slightly swollen, accompanied by a perivascular and interstitial lymphocyte-predominant infiltrate, suggesting ACR (top right panel). Hallmark features of AMR, including a more generalized swelling of the endothelial lining, thrombi in small vessels, oedema and haemorrhage as well as intravascular accumulation of macrophages, were not noted.

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REFERENCES

- [1] Sigurdardottir V, Kolsrud O, Hernandez N, Dellgren G. Endothelial cell antibody-mediated rejection and successful retransplantation in a heart transplanted patient. *Eur J Cardiothorac Surg* 2012;42:1044–6.
- [2] Reed EF, Demetris AJ, Hammond E, Itescu S, Kobashigawa JA, Reinsmoen NL *et al.* Acute antibody-mediated rejection of cardiac transplants. *J Heart Lung Transplant* 2006;25:153–9.
- [3] Berry GJ, Angelini A, Burke MM, Bruneval P, Fishbein MC, Hammond E *et al.* The ISHLT working formulation for pathologic diagnosis of antibody-mediated rejection in heart transplantation: evolution and current status (2005–2011). *J Heart Lung Transplant* 2011;30:601–11.
- [4] Uber WE, Self SE, Van Bakel AB, Pereira NL. Acute antibody mediated rejection following heart transplantation. *Am J Transplant* 2007;7:2064–74.
- [5] Kobashigawa J, Crespo-Leiro MG, Ensminger SM, Reichenspurner H, Angelini A, Berry G *et al.* Report from a consensus conference on antibody-mediated rejection in heart transplantation. *J Heart Lung Transplant* 2011;30:252–69.
- [6] Nowak G, Karrar A, Holmén C, Nava S, Uzunel M, Hulténby K *et al.* Expression of vascular endothelial growth factor receptor-2 or Tie-2 on peripheral blood cells defines functionally competent cell populations capable of reendothelialization. *Circulation* 2004;110:3699–707.
- [7] Everitt MD, Hammond MEH, Snow GL, Stehlik J, Revelo MP, Miller DV *et al.* Biopsy-diagnosed antibody-mediated rejection based on the proposed ISHLT working formulation is associated with adverse cardiovascular outcomes after pediatric heart transplant. *J Heart Lung Transplant* 2012;31:686–93.
- [8] Soares MP, Lin Y, Sato K, Stuhlmeier KM, Bach FH. Accommodation. *Immunol Today* 1999;20:434–7.
- [9] Mohacsi P, Rieben R, Sigurdsson G, Tschanz H, Schaffner T, Nydegger UE *et al.* Successful management of a B-type cardiac allograft into an O-type man with 3 1/2 year clinical follow-up. *Transplantation* 2001;72:1328–30.
- [10] Koestner SC, Kappeler A, Schaffner T, Carrel TP, Nydegger UE, Mohacsi P. Histo-blood group type change of the graft from B to O after ABO mismatched heart transplantation. *Lancet* 2004;363:1523–5.